

Transmission, Host Range and Virus-Vector Relationships of Chino del Tomate Virus (CdTV), a New Whitefly-transmitted Geminivirus of Tomato.

J. K. Brown and M. R. Nelson

SUMMARY

The transmission properties, host range, and virus-vector relationships of chino del tomate virus (CdTV), a new whitefly-transmitted geminivirus of tomato, are described. The virus is transmitted by *B. tabaci*, the sweet potato whitefly, but not by seed or sap. The virus infects members of the *Asclepiadaceae*, *Leguminosae*, *Malvaceae*, and *Solanaceae*. In virus-vector studies, minimum AAF and IAF times were 1 hour and 2 hours, respectively. The virus was retained by its whitefly vector for 4.5 and 7.3 days following 24- and 72-hr AAF respectively. Relative efficiencies of transmission for 1, 5, 10 and 20 *B. tabaci* were 15, 49, 84 and 100 percent, respectively.

The chino del tomate (CdT), or leaf curl disease of tomato (*Lycopersicon esculentum* Mill.), was first reported in cultivated tomato fields in Sinaloa, Mexico in 1970-71 (4). Presently, it occurs in tomato production areas of the west coast of Sinaloa and may affect 100 percent of the plants in a field (1). The disease is characterized by curled and rolled leaves, thickened veins, a bright-to-subdued-yellow mosaic which varies with time of the year, stunting, and a reduced fruit set (1,3). Recently, a whitefly-transmitted geminivirus, CdT virus (CdTV), was implicated as the causal agent of the disease (1,3), but information concerning the biological nature of the virus is lacking. Here, we present the results of studies involving virus transmission, experimental host range, and virus-vector relationships.

MATERIALS AND METHODS

Virus-infected leaves were collected from affected tomato plants in Culican, Sinaloa. The virus was transmitted by *B. tabaci* to healthy tomato seedlings following a 24-hr access-acquisition feed (AAF) on infected leaves and a 3-day inoculation access feed (IAF) on healthy seedlings. The resulting infected plants were used as virus source plants throughout the study.

The experimental host range was determined by inoculating seedlings of test plant species with the CdTV using viruliferous *B. tabaci* which had been reared on virus source plants. Test plant species were back-indexed to healthy 'Pole Boy' tomato indicator plants, using virus-free colony-reared whiteflies.

Mechanical inoculation experiments were done by rubbing sap from virus source plants on the cotyledons and/or true leaves of test plants. Sap was obtained by grinding symptomatic leaves in a mortar and pestle with 2 vol (w:v) 0.2M potassium phosphate buffer (pH 7.4) containing 0.5% diatomaceous earth as an abrasive.

To test for seed transmission of the virus, seeds were collected from seed pods of *D. stramonium* and *N. benthamiana*, and from the infected fruit of 'Pole Boy' tomato, all of which had been inoculated using viruliferous *B. tabaci* and maintained in the greenhouse as described above. Seeds were sown (10 seeds per pot) in 15.0-cm-diameter pots in the greenhouse, and the resulting seedlings were maintained for 8 weeks in the greenhouse for observation.

In virus-vector studies, relative efficiencies of virus transmission were determined by allowing either 1, 5, 10, or 20 whiteflies a 3-day IAF on indicator plants following a 48-hr AAF on virus source plants. The minimum AAF required for transmission was determined by allowing whiteflies access to virus source plants for either 10

min, 30 min, or 1, 2, 4, 8, 16, 24 or 48 hours, prior to transfer to indicator plants for a 3-day IAF. To determine the minimum IAF time, whiteflies were allowed either a 2-hr or 24-hr AAF on virus source plants and IAF times of 10 min, 30 min, or 1, 2, 4, 8, 16, 24 or 48 hours on indicator plants. The maximum length of virus retention (persistence) by B. tabaci was determined by allowing individual whiteflies either a 24-hr or 72-hr AAF on virus source plants, followed by serial transfer at 24-hour intervals to indicator plants for 12 consecutive days.

RESULTS

Host Range

The result of the experimental host range study and the symptoms associated with hosts are summarized in Table 1. The host range of CdTV included members within the Leguminosae, Malvaceae, and Solanaceae as well as a single species tested within the Asclepiadaceae (Table 1). Under the conditions described here, there were no CdTV hosts identified within the Amaranthaceae, Chenopodiaceae, Compositae, Cruciferae, Cucurbitaceae, Graminae, or Umbelliferae (Table 1).

Mechanical Transmission

Characteristic 'chino' symptoms did not develop in mechanically inoculated D. stramonium, N. benthamiana, or 'Pole Boy' tomato, all of which are symptomatic hosts of the virus (Table 1).

Seed transmission

Characteristic 'chino' symptoms were not observed in any of the progeny seedlings resulting from seed collected from virus-infected D. stramonium, N. benthamiana, or 'Pole Boy' tomato plants.

Virus – Vector Relations

The development of virus symptoms in indicator plants following exposure to whiteflies was considered indicative of virus transmission by B. tabaci. The results of transmission studies are reported as the mean efficiency of transmission which is based upon the means of three trials each and 15 plants per trial. The means of three trials were used to calculate the grand mean, and the standard deviation of the means is expressed parenthetically.

The relative efficiencies of virus transmission for 1, 5, 10, and 20 B. tabaci were 15 (+4.0), 49 (+14.0), 84 (+10.1), and 100% (+0.0), respectively, following a 48-hr AAF and a 3-day IAF on indicators.

The minimum AAF required for virus transmission was 1 hour after which transmission occurred 22% (+3.5) of the time. Longer AAF times of 2, 4, 8, 16, 24, and 48 hours resulted in transmission efficiencies of 26 (+6.5), 42 (+3.5), 55 (+4.04), 86 (+3.5), 91 (+10.1), and 98% (+4.0), respectively.

The minimum IAF required for virus transmission following a 2-hour AAF was 16 hours with 15% (+6.5) transmission. With a 2-hour AAF and IAF of 24-hours and 48-hours, transmission efficiencies increased to 80 (+6.5) and 98% (+4.0) respectively.

Following 24-hr and 72-hr AAF times, whiteflies retained the ability to transmit CdTV for 4.5 (+0.1) and 7.3 (+0.3) days, respectively, which is characteristic of a persistent type virus-vector relationship.

DISCUSSION

Chino del tomate or leaf curl of tomato was first recognized as a virus-like disorder in west coast Sinaloa, Mexico in 1970-71 (4) and has been a serious threat to tomato production since then (1). Symptoms in infected tomato plants are characterized by stunting, yellow foliar mottling, leaf curling and rolling, and a decrease in fruit production. Epidemics caused by CdTV are directly associated with elevated levels of whitefly vector populations which have become increasing prevalent in the Sonoran Desert and adjacent agricultural areas during the past several years. Though CdT has been reported exclusively in Sinaloa, the disease has the potential to become a serious problem in nearby tomato production areas in Mexico and the United States.

The dramatic foliar symptoms and severe reduction in fruit production associated with CdT indicate an immediate need for effective control measures on both a short and long-term basis. The direct relationship between whitefly population levels and disease incidence suggests that control of the disease could be achieved by eliminating the vector. However, attempts to adequately reduce the number of whiteflies in infected fields have

The dramatic foliar symptoms and severe reduction in fruit production associated with CdT indicate an immediate need for effective control measures on both a short and long-term basis. The direct relationship between whitefly population levels and disease incidence suggests that control of the disease could be achieved by eliminating the vector. However, attempts to adequately reduce the number of whiteflies in infected fields have generally been unsuccessful. The difficulties include placing insecticides on the lower leaf surface where whiteflies feed; a waxy covering on immature instars, which protects against insecticidal action despite adequate placement; developing resistance to insecticides; and reduced natural predator and parasite populations following insecticide application. Each of these factors is, in part, responsible for the inability to control whiteflies and thus whitefly-transmitted viruses. The implementation of crop-free periods to deprive whiteflies of over-seasoning hosts has not been feasible in the southwestern United States or Mexico to date, due to the diversity of cropping practices in most areas.

Breeding for resistance to a number of whitefly-transmitted geminiviruses of tomato is of major concern in commercial tomato production areas of the world (2). Two selections of *L. pimpinellifolium* LA121 and LA1478, which were reported to be highly resistant to a related virus, TYLCV in the Middle East and Sudan exhibited tolerance to CdTV infection in greenhouse studies (Table 1). These results indicate the potential feasibility of developing tolerant or resistant tomato selections.

REFERENCES

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4. Gallegos, H. M. L. 1978. *Enfermedades de cultivos en el estado de Sinaloa*. SARH, Sinaloa, Mexico. 213 pp.

Table 1. Results of a host range study of Chino del tomate virus (CdTV) by whitefly transmission using a 48-hr acquisition-access feeding on virus source plants, a 3-day inoculation-access feeding on test plants, and back-indexing to 'Pole Boy' tomato indicator plants

TEST PLANT	SYMPTOMS ^a / RESULTS OF BACK - INDEXING ^b	TEST PLANT	SYMPTOMS ^a / RESULTS OF BACK - INDEXING ^b
Amaranthaceae		Malvaceae (cont.)	
<i>Gomphrena globosa</i> L.	NS/-	<i>Malva micaensis</i> All.	M,Mo,LC/+
Asclepiadaceae		<i>Malva parviflora</i> L.	S,Mo,LC/+
<i>Cynanchum acutum</i> L.	M,VC/+	<i>Sida</i> sp. Gray	NS/-
Chenopodiaceae		Solanaceae	
<i>Beta vulgaris</i> L. 'H-9'	NS/-	<i>Capsicum annuum</i> L.	
<i>Chenopodium album</i> L.	NS/-	'Anaheim'	M,Mo,VC/+
Compositae		<i>C. frutescens</i> L. 'Tabasco'	M,Mo,VC/+
<i>Lactuca sativa</i> L. 'Salina'	NS/-	<i>Datura stramonium</i> L.	S,Mo,LC/+
<i>Zinnia elegans</i> Jacq. 'Lilliput'	NS/-	<i>D. tatula</i> L.	M,Mo,LC/+
Cruciferae		<i>Lycopersicon esculentum</i>	
<i>Capsella bursa-pastoris</i> L. Medic.		Mill. 'Pole Boy'	S,Mo,LC/+
<i>Rhaphanus sativus</i> L. 'Coment'		<i>L. hirsutum</i> Mill. LA1777	M,VC/+
Cucurbitaceae		<i>L. pennellii</i> (Correll)	M,VC/NT
<i>Citrullus vulgaris</i> Schrad	NS/-	D'Arcy LA716 ATICO	
'Charleston Gray'		<i>L. peruvianum</i> (L.)	
<i>Cucumis melo</i> L. 'Topmark'	NS/-	Mill. LA111	M,Mo/+
<i>C. sativus</i> L. 'Bush Champion'	NS/-	<i>L. peruvianum</i> (L.) Mill.	
<i>Cucurbita maxima</i> Duch. 'Big Max'	NS/-	PI126935	M,Mo/+
<i>C. pepo</i> L. 'Early Acorn'	NS/-	<i>L. peruvianum</i> (L.) var.	
Graminae		<i>glandulosum</i> LA1292	M,Mo/+
<i>Zea Mays</i> L. 'Golden X Bantam'		<i>L. pimpinellifolium</i>	
Leguminosae		(Jusl.) Mill. LA1478	NS/+
<i>Cicer arietinum</i> L. 'Kabuli Type'	NS/-	<i>L. pimpinellifolium</i>	
<i>Lens culinaris</i> Medic. 'Chilean		(Jusl.) Mill. LA722	M,Mo/+
Lentil 78'	M,Mo,VC/+	<i>L. pimpinellifolium</i>	
<i>Phaseolus aureus</i> Roxb.	M,Mo,+	(Jusl.) Mill. LA121	M,Mo/+
<i>Phaseolus vulgaris</i> L. 'Red Kidney'	M,LC,Mo/+	<i>Nicotiana benthamiana</i> L.	S,Mo,LC/+
<i>Pisum sativum</i> L.	NS/-	<i>N. clevelandii</i> Gray	S,Mo,LC/+
<i>Vicia faba</i> L.	NS/-	<i>N. glutinosa</i> L.	M,LC/+
<i>Vigna unguiculata</i> subsp.		<i>N. repanda</i> L.	M,LC/+
unguiculata (L.) Walp.		<i>N. rustica</i> L.	M,Mo,LC/+
'California Blackeye'	NS/-	<i>N. tabacum</i> L. 'Samsun'	M,Mo,LC/+
Malvaceae		<i>N. tabacum</i> L. 'Xanthi'	M,LC/+
<i>Althaea rosea</i> Car.		<i>N. tabacum</i> L.	
'Chater's Double Mix'	NS/-	'White Burley'	NS/+
<i>Gossypium hirsutum</i> L.		<i>Solanum melongena</i> var	
'Delta Pine 70'	NS/-	<i>esculentum</i> Nees.	
<i>Hibiscus esculentus</i> L.		'Black Beauty'	NS/-
'Clemson Spineless'	NS/-	<i>S. tuberosum</i> L.	
		'White Pontiac'	NS/-
		<i>Physalis peruviana</i> L.	NS/-
		Umbelliferae	
		<i>Daucus carota</i> L. var	
		<i>sativa</i> 'Danvers Half	NS/-
		Long'	

^a NS = no symptoms; M = mild symptoms; S = severe symptoms; LC = Leaf curling, Mo = Mottle or mosaic, VC = vein clearing.

^b + = host, - = nonhost, and NT = not tested in back-indexing.